THE LINKAGE OF TWO FACTORS IN DROSOPHILA THAT ARE NOT SEX-LINKED.

T. H. MORGAN AND CLARA J. LYNCH.

In addition to the eight sex-linked factors described for *Drosophila*, a number of other factors have been found that are not sex-linked. They are now being studied in order to discover which, if any, of them are linked to each other. Two that have been found to be linked are here described. These are the yellow

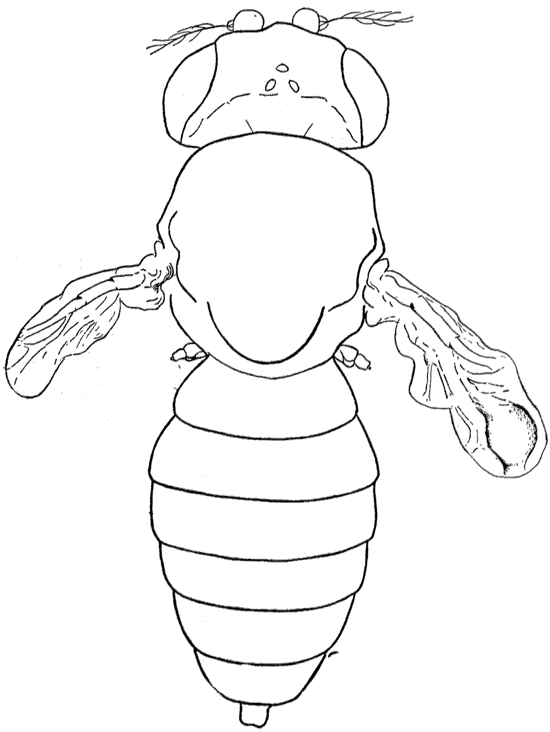

FIG. A.

factor in the absence of which the flies are black, and a factor (in the complex of factors that produces the normal wing) in the absence of which the fly is wingless.

In previous papers on *Drosophila* a large amount of data has been given showing that in regard to any sex-linked factor, the, yellow factor, *Y*, follows Mendel's law of independent segregation; in other words, the yellow factor segregates independently of all

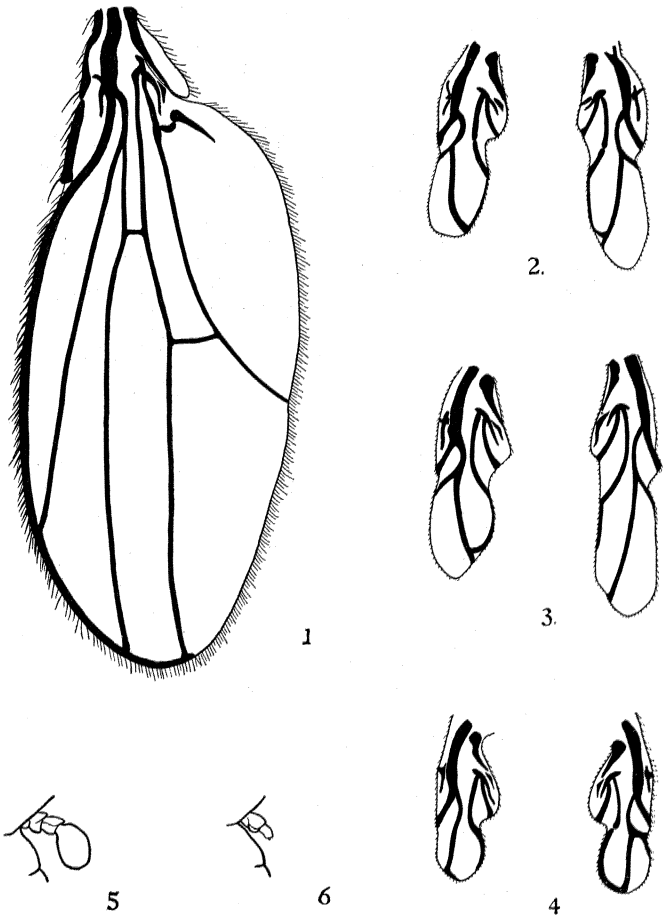

FIG. B.

sex-linked factors. The fact that wingless flies do not show sex-linked inheritance has also been reported, but the character of the wing itself and its mode of origin have not been fully described.

The size in relation to the body, and general character of the wings are shown in Fig. A. They are little more than scales held

out at right angles at the sides of the body. The details of the venation are shown in the three pairs of wings shown in Figs. B, 2, 3 and 4, while for comparison a normal ("long") wing drawn to scale is shown in Fig. 1.

The wingless wing represents in the main the basal portion of the normal wing. The marginal vein is missing, but the cross vein connecting the third and fourth longitudinal veins is often present as an apparently marginal vein. The basal parts of all five longitudinal veins are present.

The balancers are much reduced in size and modified in character. A normal balancer is shown in Fig. 5 and one from a wingless fly in Fig. 6. The terminal segment which is large in the wild fly is represented by only a trace and the second segment is reduced, but the basal segment approaches in size that of the wild type.

The wingless flies appeared in the "truncate" stock and represented at first the wingless condition of the truncate type of wing; owing to the low viability of the truncate flies the winglessness was transferred to normal stock, but as it is difficult or impossible to distinguish between Wingless of truncate stock and wingless of long-winged stock the material was not at first quite homogeneous except in so far as being wingless. There was also present at first in the wingless stock another character, namely, balloon wings, which appeared at about the same time as Wingless in the truncate stock and proved as difficult to separate from the Wingless as Truncate itself. As a result, balloon wings appear in the first cross given below. The cross was made over a year ago by Miss E. M. Wallace.

$$\begin{array}{ccccc} P_1 & & F_1 & & F_2 \\ \dfrac{\text{Wingless } \male}{\text{Winged } \female} & = & \dfrac{\text{Winged } \male}{\text{Winged } \female} & = & \left\{\begin{array}{lr} \text{Winged } \female, & 1{,}136 \\ \text{Winged } \male, & 1{,}038 \\ \text{Wingless } \female, & 259 \\ \text{Wingless } \male, & 236 \\ \text{Balloon } \female, & 12 \\ \text{Balloon } \male, & 3 \end{array}\right. \end{array}$$

The sum of the winged and balloon is 2,189 and that of the wingless is 495, which gives a ratio of 4.42 to 1. It is clear that

winglessness is recessive to normal, that it is not sex-linked, and that it has a lower viability than has the normal. The reciprocal cross gave:

$$\begin{array}{c} P_1 \\ \text{Winged ♂} \\ \text{Wingless ♀} \end{array} = \begin{array}{c} F_1 \\ \text{Winged ♀} \\ \text{Winged ♂} \end{array} = \overset{F_2}{\left\{ \begin{array}{lr} \text{Winged ♀,} & 694 \\ \text{Winged ♂,} & 639 \\ \text{Wingless ♀,} & 146 \\ \text{Wingless ♂,} & 127 \\ \text{Balloon ♀,} & 4 \\ \text{Balloon ♂,} & 1 \\ \text{Miniature ♀,} & 1 \\ \text{Miniature ♂,} & 24 \end{array} \right.}$$

One at least of the grandparental wingless flies must have been responsible for the miniatures, but the numbers are too small to affect the other ratio seriously. There were 1,363 long (including balloon and miniature) flies, and 273 wingless, or 4.9 to 1.

In a recent cross between long-winged ♀ and wingless ♂ there were present in the F_2 generation 1,081 long winged F_2 flies, and 280 Wingless; a ratio of 3.9 to 1. The reciprocal cross gave in F_2 1,213 long-winged flies and 273 Wingless or a ratio of 4.4 to 1. Compared with the preceding the results show that the viability of the Wingless has not changed. It is curious to find that the F_2 ratio in this cross and its reciprocal differ in both instances in the same direction as in the earlier cross, but this may be only a coincidence.

Linkage of the Yellow Factor, *Y*, and the Wing Factor, *W*.

The linkage in question is most simply shown in the following result: wingless Grays were mated to long-winged Blacks, and gave in F_1 long-winged gray males and females. These inbred gave in the F_2 generation:

Winged Gray.		Winged Black.		Wingless Gray.		Wingless Black.	
♂	♀	♂	♀	♂	♀	♂	♀
854	1,004	410	506	226	329	0	0

There are no wingless black flies in the F_2 generation, which the Mendelian expectation calls for. Their absence can only be

explained by strong linkage of the yellow factor and the factor for wings. The Mendelian expectation (unmodified by linkage and viability) is three long Grays to one long Black (3 : 1). *There are actually 1,858 long gray flies to 916 long black, or a ratio of 2 to 1. This is the linkage ratio when two strongly or completely linked factors are concerned.*

Since the males and females count equally in the result it is not necessary to separate them. We are indebted to Mr. A. H. Sturtevant for the following F_2 count derived from the same F_1 material as the last. We are also indebted to him for being first in pointing out the interpretation of this result.

Winged Gray.	Winged Black.	Wingless Gray.	Wingless Black.
458	230	185	0

There are 458 winged Grays to 230 winged Blacks, or almost exactly 2 : 1.

In the following analysis Y = yellow factor; y its absence; B = black factor (sex-linked), and b its absence. The formula for the female gray fly is $YBXYBX$, and for the male, $YBXYb$. The formula for the female black fly is $yBXyBX$, and for the male is $yBXyb$.

The evidence indicates that the wingless condition differs from the winged condition by a loss. Large W may then stand for that "factor" in the original complex that has been lost. In this sense large W means in this case Wings (here long); *and small w stands for this loss and in the formulæ represents the wingless condition, which is of course the result of the interaction of all the remaining wing factors.*

	$wYBX$—$wYBX$	Wingless gray ♀.
	$WyBX$—Wyb	Winged black ♂.
F_1	$wYBX\ WyBX$	Winged gray ♀.
	$wYBX\ Wyb$	Winged gray ♂.
Gametes of	$wYBX$—$WyBX$	Eggs.
F_1	$WyBX$—$wYBX$—Wyb—wYb	Sperm.
	$wYBX\ WyBX$	Winged gray ♀.
	$wYBX\ wYBX$	Wingless gray ♀.
	$WyBX\ WyBX$	Winged black ♀.
	$WyBX\ wYBX$	Winged gray ♀.

wYBX Wyb	Winged gray ♂.
wYBX wYb	Wingless gray ♂.
WyBX Wyb	Winged black ♂.
WyBX wYb	Winged gray ♂.

In the F_1 female, gametes having the composition *WYBX* and *wyBX* would be expected on any assumption except that of strong linkage of *wY* and *Wy*, and likewise in the male, the gametes *WYBX*, *wyBX*, *WYb* and *wyb* would be expected. For the sake of convenience I have omitted to represent the cross-over gametes in the above analysis.

In another cross, miniature-winged, black, white-eyed flies were mated to wingless, gray, red-eyed flies. The result is complex, but only in the sense of giving in one cross 20, F_2, classes, and in the reciprocal cross 13, F_2, classes. The results, however, as far as linkage is concerned, are perfectly evident. The fact is also brought out that the factor for Wings shows no linkage to the sex-linked factor (*C*) for eye color, and to (*R*) the factor in the absence of which the wings are miniature.

Miniature White Bl ♂ by Wingless Red Gray ♀

GR Long ♂ and ♀

F_2 Females

GRL	*BlRL*	*GR* Wingless
532	268	235

Males

GRL	*GWhL*	*BlRL*	*BlWhL*	*GRM*	*GWhM*	*BlRM*	*BlWhM*	*GRW-g-s*	*GWhW-g-s*
173	49	81	38	92	165	30	52	117	90

There are three classes of females and ten classes of males. The wingless flies only occur as Gray—no black wingless flies being present. The alteration in the ratios, due to the linkage between the two sex-linked factors *C* and *R*, is neglected here. The analysis follows:

	wRMCBYX—wRMCBYX	Wingless red gray ♀.
	WrMcByX—Wrmcby	Min. white black ♂.
F_1	*wRMCBYXWrMcByX*	Winged gray red ♀.
	wRMCBYXWrmcby	Winged gray red ♂.

Gam-	*WRMCByX–wRMCBYX–WrMcByX–wrMcBYX*	eggs
etes	*WRMcByX–wRMcBYX–WrMCByX–wrMCBYX*	
of F_1	*WRMCByX—wRMCBYX—Wrmcby—wrmcbY*	Sperm.

Females.

WRMCByXWRMCByX	Long red black.
WRMCByXwRMCBYX	Long red gray.
WRMcByXWRMCByX	Long red black.
WRMcByXwRMCBYX	Long red gray.
wRMCBYXWRMCByX	Long red gray.
wRMCBYXwRMCBYX	Wingless red gray.
wRMcBYXWRMCByX	Long red gray.
wRMcBYXwRMCBYX	Wingless red gray.
WrMcByXWRMCByX	Long red black.
WrMcByXwRMCBYX	Long red gray.
WrMCByXWRMCByX	Long red black.
WrMCByXwRMCBYX	Long red gray.
wrMcBYXWRMCByX	Long red gray.
wrMcBYXwRMCBYX	Wingless red gray.
wrMCBYXWRMCByX	Long red gray.
wrMCBYXwRMCBYX	Wingless red gray.

Males.

WRMCByXWrmcby	Long red black.
WRMCByXwrmcbY	Long red gray.
WRMcByXWrmcby	Long white black.
WRMcByXwrmcbY	Long white gray.
wRMCBYXWrmcby	Long red gray.
wRMCBYXwrmcbY	Wingless red gray.
wRMcBYXWrmcby	Long white gray.
wRMcBYXwrmcbY	Wingless white gray.
WrMcByXWrmcby	Min. white black.
WrMcByXwrmcbY	Min. white gray.
WrMCByXWrmcby	Min. red black.
WrMCByXwrmcbY	Min. red gray.
wrMcBYXWrmcby	Min. white gray.
wrMcBYXwrmcbY	Wingless white gray.
wrMCBYXWrmcby	Min. red gray.
wrMCBYXwrmcbY	Wingless red gray.

The reciprocal cross gave the following results.

Minature White Bl ♀ by Wingless Red Gray ♂

Gray Red ♀ and Miniature Gray White ♂

Females.

GRL	*GWhL*	*BlRL*	*BlWhL*	*GRM*	*GWhM*	*BlRM*	*BlWhM*	*GRW-g-s*	*GWhW-g-s*
220	45	88	45	96	197	46	62	113	95
468				401				208	

Males.

GRL	*GWhL*	*BlRL*	*BlWhL*	*GRM*	*GWhM*	*BlRM*	*BWhM*	*GRW-g-s*	*GWhW-g-s*
225	96	120	35	92	201	41	99	131	105
473				433				236	

In this reciprocal cross there are ten classes each of females and males. Each wingless class is made up (as in the other cross also) of wingless long and wingless miniature. These, if they could be separated, would give, in all, twelve classes of males and females.

The analysis of this cross is similar in principle to that given above and can readily be reproduced by the method used for that cross. How closely the yellow factor and that for Wing are united will be determined later by suitable tests.

On the assumption that factors are contained in chromosomes, it will be apparent that this new case of linkage means that both *W* and *Y* are contained in a chromosome different from the sex chromosome.

After the foregoing results were completed, a doubt arose as to whether we could with certainty detect black wingless flies, even if present, since they would lack one of the most characteristic features of black flies, namely, the wings. We determined to test some of the F_2 flies by mating to long-winged black flies, for we thought, at that time, that this would be a legitimate test for the presence of black flies amongst the wingless offspring. But while it is true that the appearance of black offspring would be expected, if black flies were present, nevertheless the appearance of such flies does not in itself show that black flies were actually present amongst the flies tested; for, as the following analysis will show, crossing over may have occurred when the gametes were produced, and yet no F_2 black

wingless flies result. The test showed, in fact, that crossing over had occurred, but the result was obscured, because, necessarily, the F_2 flies would still be gray, unless the crossing over had occurred simultaneously in both sexes and the cross-over gametes happened to meet. The following experiments will illustrate this point:

Seven F_2 wingless males were paired with 15 long-winged black females, and gave 133 gray females, and 96 gray males.

But five wingless females mated to fifteen long black males gave: Gray, ♀, 64; ♂, 61; Black, ♀, 8; ♂, 7. The last result shows that crossing over had occurred in the gametes of one F_1 fly, which gave rise to one fly (at least) that was heterozygous for color, and from this fly came the black flies that appeared in the test. The point of particular interest is that this crossing over had not been apparent in the F_2 gray flies, and the reason for this is not far to seek. If crossing over between wY and Wy in the gametes of the F_1 generation had occurred, the gametes would be: WYB, wYB, WyB, wyB.

If this fly were fertilized by an ordinary F_1 male, whose gametes would be wYB and WyB, all the wingless flies would be gray, and those grays derived from the crossing-over (however rare their occurrence) would be heterozygous for color. Only a back cross by black flies would make evident in the next generation that crossing-over had really occurred. How often it occurs remains to be discovered by further experiments. The ratio of 2 to 1 in the F_2 gray flies shows, however, that crossing-over is not common, and the absence of black wingless flies in the F_2 generation indicates this even better. For, if crossing over were common the cross-over gametes of the F_1 flies would occur often enough for like to meet like and produce black wingless flies.